Commonwealth of Virginia
Department of General Services
Division of Consolidated Laboratory Services
Richmond, Virginia

Carbapenem Resistant Enterobacteriaceae (CRE) and Pseudomonas aeruginosa (CRPA) Testing Instructions

I. CURRENT DCLS LABORATORY TESTING

Test Type	Method
Identification Confirmation	MALDI-TOF (Bruker Biotyper)
Antimicrobial Susceptibility Testing (AST)*	Broth Microdilution/Sensititre
Phenotypic testing - Carbapenemase Production	Modified Carbapenem Inactivation Method (mCIM)
Resistance Mechanism Identification	Real-Time PCR

*** Submitting laboratories are REQUIRED to provide AST results when submitting suspected CRE/CRPA isolates to DCLS for testing. These AST results are assessed upon receipt to determine potential pan-resistance.**

TESTING METHODS

- **mCIM** – Phenotypic testing for carbapenemase production will be performed for all CRE and CRPA isolates.
- **AST** – The following drug Minimum Inhibitory Concentration (MIC) values will be determined for all CRE isolates and mCIM-positive CRPA isolates:
  1. Amikacin
  2. **Aztreonam***
  3. **Cefepime***
  4. **Cefotaxime*** (CRE only)
  5. **Ceftazidime***
  6. Ciprofloxacin
  7. **Colistin***
  8. Ertapenem (CRE only)
  9. **Imipenem***
  10. Levofloxacin
  11. **Meropenem***
  12. **Piperacillin/Tazobactam****
  13. Polymyxin B
  14. Tobramycin
  15. Trimethoprim/Sulfamethoxazole (CRE only)

*** Testing required by the CDC's Antibiotic Resistance Laboratory Network (ARLN) for CRE and CRPA isolates.**
**** Testing required by the CDC's ARLN for *Pseudomonas aeruginosa* isolates.**

- **Real-time PCR** - Analyses will be performed on the following resistance marker genes for all CRE isolates and mCIM-positive CRPA isolates:
  1. KPC
  2. NDM
  3. OXA-48-like
  4. VIM
  5. IMP
  6. *mcr*-1 and *mcr*-2

II. SPECIMEN TYPES

- Pure suspect CRE isolates on slant or plate media.
  1) Member of the Enterobacteriaceae family
  2) Resistance to at least 1 carbapenem:
     - imipenem, meropenem or doripenem = **MIC of ≥4 µg/mL**
     - ertapenem = **MIC of ≥2 µg/mL**

Note: *Morganella spp.*, *Proteus spp.*, and *Providencia spp.* have intrinsic elevated MICs to imipenem. MIC results for meropenem, doripenem, and/or ertapenem must be used to determine if these organisms should be forwarded to DCLS for testing.

- Pure suspect CRPA isolates on slant or plate media.
  1) Isolate identified as *Pseudomonas aeruginosa*
  2) Resistance to at least 1 carbapenem:
     - imipenem, meropenem or doripenem = **MIC of ≥8 µg/mL**
  3) Mucoid *Pseudomonas aeruginosa* isolates will NOT be tested

III. SPECIMEN TRANSPORT

- Ship isolates at room temperature.
- Submit a completed DCLS Clinical Microbiology/Virology Test Request form with isolate, to include the following:
    1) Complete patient information (including address)
    2) Complete submitter information
    3) Date of specimen collection
    4) Specimen source
    5) Organism identification, denoting "suspected carbapenem resistance"
    6) Outbreak ID number (if applicable)
- Include copy of laboratory's AST results in specimen package
- **Shipping Address:**

  Division of Consolidated Laboratory Services
  Attn: CRE Team
  600 North 5th Street
  Richmond, Virginia 23219

IV. RESULTS REPORTING

- Turnaround time for reporting results will be within 6 business days of specimen receipt.
- Alert value results (**results requiring additional reference laboratory testing**) will be verbally reported within 1 working days of results. An alert email notification will be sent to the CDC as well.
- Non-alert-value, positive results (**results NOT requiring additional reference laboratory testing**) will be verbally reported to the submitting laboratory and VDH within 2 working days of results.
- Negative results will not be verbally reported.
- A hard copy report of final test results will be provided to the submitter by mail.
    - CRE final results will include: identification, AST, phenotypic, and molecular
    - CRPA final results will include: if mCIM negative, identification and phenotypic only; if mCIM positive, identification, AST, phenotypic, and molecular
- VDH will receive electronic reporting of results.
- Reference laboratory results for additional characterization testing will be provided to the submitting laboratory and VDH within 2 working days of receipt from the reference laboratory.

V. CONTACT INFORMATION

- **For questions regarding testing, specimen collection or shipment, please contact one of the following:**
    - Dr. LaToya Griffin-Thomas (804-648-4480, ext. 281)
    - Microbial Reference Laboratory (804-648-4480, ext. 345 or 251)
    - Molecular Detection and Characterization Laboratory (804-648-4480, ext. 298)
    - DCLS 24/7 Emergency Mobile Number (804-335-4617)

VI. RESOURCES

Updated information on CRE/CRPA testing can be found on the DCLS website, under the "Hot Topics" link:
https://dgs.virginia.gov/division-of-consolidated-laboratory-services/updates/hot-topics/

Link to DCLS Clinical Microbiology/Virology Test Request Form:
https://dgs.virginia.gov/globalassets/business-units/dcls/documents/sample-kits/dcls-micro_viro-form_fillable_rw.pdf